Two tightly-linked genes controlling grain length underlie a major grain weight QTL in polyploid wheat

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Final grain yield is an important, but complex trait

- Number of spikes
- Number of grains
- Grain weight
Dissecting Grain Size: **Grain Length** and **Grain Width**

- **Grain weight**
- **Grain Size**
  - **Grain Length**
  - **Grain Width**
- **Grain Filling**
Reducing complexity using Near Isogenic Lines (NILs)

Doubled Haploid population
Charger x Badger

Inner tracks
SNP variation (iSelect 90k chip)
1. Parental SNPs
2. BC₄ NILs SNPs

Outer track
Combined LOD score
(12 environments)

Brinton et al. bioRxiv 2017
5A+ NILs have increased TGW

Brinton et al. bioRxiv 2017
5A+ NILs have increased TGW

Grain length is the main driver of increased TGW

Brinton et al. bioRxiv 2017
5A QTL increases grain length uniformly

Brinton et al. *bioRxiv* 2017
Longer cells or more cells?

5A -
5A +

Cell expansion

Cell division

Brinton et al. bioRxiv 2017
5A+ grains have increased cell length

Per genotype group:
Grains measured = 24
Cells measured = >1500

Brinton et al. bioRxiv 2017
5A+ grains have increased cell length

Brinton et al. bioRxiv 2017
5A+ grains have increased cell length *independent of absolute grain size*. 

Brinton et al. *bioRxiv* 2017
When does the 5A QTL act?

5A QTL acts during grain development

Using RNAseq: differential expression between NILs

50 transcripts differentially expressed
20% of DE genes relate to ubiquitin & protein turnover
20% of DE genes relate to ubiquitin & protein turnover

GW2: RING-type E3 ligase

GW2
gw2
Song et al Nature 2007

DA2
da2
Xia et al Plant Cell 2013

GW2-A
gw2-A
GW2-A
gw2-A
Simmonds et al TAG 2016

20% of DE genes relate to ubiquitin & protein turnover
Fine mapping of the 5A QTL for grain length

Brinton et al. bioRxiv 2017
Fine mapping of the 5A QTL for grain length

Grain length (mm)
Fine mapping of the 5A QTL for grain length

- **New Rec group 1:**
  - 5A-: n=33
  - 5A+: n=3

- **New Rec group 2:**
  - 5A-: n=3
  - 5A+: n=3

- **New Rec group 3:**
  - 5A-: n=13
  - 5A+: n=21

- **New Rec group 4:**
  - 5A-: n=21
  - 5A+: n=4

- **New Rec group 5:**
  - 5A-: n=3
  - 5A+: n=3

- **New Rec group 6:**
  - 5A-: n=27
  - 5A+: n=6

- **Grain length (mm):**
  - 5A-:
    - 1.1: n=27
    - 1.2: n=6
  - 5A+:
    - 1.1: n=27
    - 1.2: n=6
Fine mapping of the 5A QTL for grain length

New Rec group

5A- 5A+
1 1
2 2
3 3
4 4
5 5
6 6

n=33 n=3
n=3 n=3
n=13 n=21
n=4 n=21
n=3 n=3

Grain length (mm)

New Rec group

1 1
1.1 1.2

n=27 n=6
n=3 n=3

Grain length (mm)
Fine mapping of the 5A QTL for grain length

Grain length (mm)

- New Rec group
  - 1
  - 2
  - 3
  - 4
  - 5
  - 6

- 5A-
- 5A+

- n=33
- n=3
- n=13
- n=21
- n=4
- n=21

- 5A-
- 5A+

- n=27
- n=6
- n=10
- n=3

Mean length

- 6.4
- 6.5
- 6.6
- 6.7
- 6.8
- 6.9
Fine mapping of the 5A QTL for grain length

Grain length (mm)

5A- 5A+

New Rec group

n=33 n=3
n=33 n=3
n=13 n=10
n=21 n=3
n=4 n=3
n=21 n=3

n=27 n=6
n=10 n=3

5A- 5A+

1 1.1
2 1.2
3 3.1
4 3.2
5
6

100 Mbp 500 Mbp

5A- 5A+

n=3 n=3
n=21 n=21
n=4 n=4
n=21 n=13

n=33 n=33
n=13 n=13
n=4 n=4
n=21 n=21

n=27 n=27
n=10 n=10
n=3 n=3

Grain length (mm)

n=3 n=3
n=10 n=10
n=3 n=3

n=21 n=21
n=4 n=4
n=21 n=21

n=27 n=27
n=10 n=10
n=3 n=3

Grain length (mm)
Two tightly linked genes underlie the 5A QTL
GL-A1 and GL-A2 are not fixed in UK germplasm
**GL-A1 and GL-A2 are not fixed in UK germplasm**

- 5A- Charger
  - Short grain
  - Dickens
  - Panorama
  - Solstice
  - Relay
  - Santiago
  - Claire

- 5A+ Badger
  - Long grain

- GL-A1

- Cadenza

- Relay

- Santiago

- Claire

- GL-A2
Two tightly linked genes underlie the 5A QTL. GL-A1 and GL-A2 are not fixed in UK germplasm.
Testing candidates using exome-sequenced TILLING mutants

15 candidate genes

A genome mutant  x  B genome mutant  →  Kronos TILLING mutants (AB)

www.wheat-tilling.com
Krasileva et al. PNAS 2017
Can we combine different grain size genes?

Cell expansion +5%
Cell division +5%

Cell expansion +10% ??
Cell division +10% ??
Can we combine different grain size genes?

Grain length (5A QTL)

- Cell expansion
- Cell division

Cell expansion

Cell division

+10% ??

+5%
Can we combine different grain size genes?

Simmonds et al. *BMC Plant Biol* 2014
Can we combine different grain size genes?

- **Cell expansion**
  - 5A -
  - 5A +

- **Grain length (5A QTL)**

- **Cell division**
  - 6A -
  - 6A +

- **Grain width (6A QTL)**

Simmonds et al *BMC Plant Biol* 2014

**Graph:**
- Thousand grain weight (g)
- BC$_4$ Paragon NILs 2016

- -/-
- 6A+
- 5A+
- 5A+ 6A+
Understanding the genes and mechanisms controlling grain size will allow us to increase final yield in wheat.